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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 : A61K 35/78	A2	(11) International Publication Number: WO 00/00211
		(43) International Publication Date: 6 January 2000 (06.01.00)

(21) International Application Number: PCT/IL99/00355

(22) International Filing Date: 28 June 1999 (28.06.99)

(30) Priority Data:
125138 29 June 1998 (29.06.98) IL

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(81) Designated States: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
Without international search report and to be republished upon receipt of that report.

(54) Title: HYDROCOLLOIDS OBTAINED FROM PORTULACA OLERACEA AND THE USE THEREOF

(57) Abstract

The present invention relates to a hydrocolloid obtained by extraction from Portulaca Oleracea having the composition as shown in Table I in which the sugar contents in the fibers are as shown in Table II and the composition of the ash portion is as given in Table III.

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HYDROCOLLOIDS OBTAINED FROM PORTULACA OLERACEA AND THE USE THEREOF

The present invention relates to hydrocolloids obtained from Portulaca Oleracea, (hereinafter "the plant") and to the use thereof.

The plant is known as is the composition thereof. (See Ali I Mohamed et. al.; Chemical Composition of Portulaca Oleraca; Plant Foods for Human Nutrition, 45, 1994, pp. 1-9).

It is known that the plant comprises 3.5-4.5% of a hydrocolloid which is partially soluble in water. However, no use of said hydrocolloids is known so far.

The hydrocolloid may be obtained by extracting same from the plant by a method which is substantially described in G. Wenzel et. al.; The Viscous Mucilage from the Weed Portulaca Oleracea, L; Applied Biochemistry and Biotechnology, Vol. 24/25, 1990, pp. 341-353. Said authors analyzed the hydrocolloid to a certain extent but did not analyze all parts of same. The results of the analysis performed by the present inventors are shown in Table I.

Table I

Component	Average Content (%)
Humidity	10 - 11
Fat	0.5 - 0.6
Proteins	2.5 - 3.2
Nutritionable water soluble fibres	60 - 62
Nutritional non water soluble fibres	7.5 - 8.5
Total nutritional fibres	68 - 70

The hydrocolloid was extracted in accordance with the method described by G. Wenzel et. al. The white powder obtained was analyzed, in particular the sugar contents of the fibers; and the ash portion of the hydrocolloid.

Said analysis gave the following results:

The sugar contents of the fibers are shown in Table II

Table II

D-galactose	L-arabinose	L-rhamnose	D-xylose	D-galacturonic acid
40	: 20	: 5	: 1	: 31

Said values are substantially within the limits of the results given by Wenzel et. al.

The analysis of the ash portion which had so far not been performed gave the results shown in Table III.

Table III

Contents mg/kg	
1,400	Ca
20,000	Mg
60,000	K
2,000	Na
900	Fe
460	S
720	P
60	Al

The presence of transition and heavy metals in the ash portion was also determined and it was found that they were present, if at all, in very small portions. Therefore, the presence of said transitional metals and/or heavy metals may be disregarded.

The present invention thus consists in a hydrocolloid obtained by extraction from Portulaca Oleracea having the composition shown in Table I in which the sugar contents in the fibres are as shown in Table II and the composition of the ash portion is as given in Table III.

The specific characteristic features regarding the molecular

weight, the solubility in water, the viscosity, the surface activity and the interface activity of the hydrocolloid were also determined.

It has been found that the molecular weight of the fraction of the hydrocolloid has a distribution as shown in Table IV.

Table IV

34%	up to 10^4 daltons
12%	between 10^4 - 10^5 daltons
33%	between 10^5 - 10^7 daltons
11%	between 10^7 - 10^8 daltons
10%	over 10^8 daltons

It has been found that the solubility of said hydrocolloid in water is as shown in Table V.

Table V

Concentration upto 0.8%	well soluble, gives a nearly clear solution
Concentration upto 1.2%	well dispersible, a white solution
Concentration upto 2.0%	the aqueous phase becomes viscous, a white-gray porri-dge
Concentration of 2.0% upto 6.0%	the viscosity is gradually increased, the hydrocolloid becomes harder and it is less soluble in water
Concentration over 6.0%	two phases are obtained; the upper one is viscous; the lower one a gelatine like solid.

It has been found that the viscosity of said hydrocolloid is as shown in Table VI.

Table VI

Conc. wt%	0.05	0.1	0.2	0.3	0.5	0.7
cst = centistoke	1.46	2.00	2.99	4.08	6.77	9.94

Said data show that said hydrocolloids are not very viscous, which means that one may obtain a relatively concentrated solution which cannot be obtained with other hydrocolloids.

Moreover, said hydrocolloid has a good surface tension. Thus, e.g it may reduce the surface tension of water in a concentration of 0.6% of the weight of the material, from 72 dyn/cm to 47 dyn/cm.

The interface tension between tetra-decane and water is in general 44 dyn/cm and in the presence of the hydrocolloid according to the present invention it is reduced to 18 dyn/cm.

The ratio between the concentration of the hydrocolloid and the surface tension and of the interface tension is shown in Table VII.

Table VII

Conc. hydrocolloid dyn/cm	Surf. Tension Air/water dyn/cm	Interface Tension water/oil wt%
0.075	49.8	-
0.1	49.5	23.0
0.2	48.4	21.0
0.4	47.6	19.1
0.6	47.0	18.7
0.8	47.0	17.8

All the above data regarding the molecular weight, the solubility in water, the viscosity, the surface tension and the interface tension of the hydrocolloid according to the present invention have so far not been known.

It has surprisingly been found that the hydrocolloid according to the present invention may be used, inter alia, in the following ways:

1. It may be part of an emulsion;
2. It can be part of a pharmaceutical preparation which may be used, e.g. for
   a. reducing the sugar level in the blood; and
   b. reducing the blood pressure.

The present invention thus consists also in emulsions comprising as active ingredient a hydrocolloid according to the present invention.

A preferred emulsion is that of , e.g. a paraffin oil, such as tetra-decane, which is dispersed in an aqueous solution of the hydrocolloid.

However, the present invention is not restricted to the above emulsions. Thus similar emulsions may be prepared with other oils, e.g. soya oil, olive oil, coconut oil, castor oil, tricaprylin oil, sunflower oil, etc.

A stable emulsion can also be obtained when toluene is the oil phase.

When the hydrocolloid solution is further purified by centrifugation the fraction having a higher molecular weight is precipitated. The fraction remaining in the solution enables the preparation of emulsions which are stable for a long period having comprising drops having a much lower average size.

The lower the pH of the hydrocolloid dispersion a more stable emulsion is being obtained having a lower average drop size (1-2 micrometers).

The reduction of the ion strength also causes an increase of

the stability of the emulsion.

The hydrocolloid dispersion which is prepared at room temperature also yields a more stable emulsion than those prepared at other temperatures.

The stability of the emulsion according to the present invention may be increased by the addition of another suitable emulsifier, e.g Tween 20; sorbitan monolaurate-20-ethoxylate; sucrose esters; etc.

The present invention also consists in pharmaceutical preparations comprising as active ingredient a hydrocolloid according to the present invention. Said pharmaceutical preparation is also used in particular for reducing the sugar content in the blood and/or for reducing the blood pressure.

The pharmaceutical composition may be an emulsion according to the present invention. However, it may be any suitable tablet, capsule, solution, etc. comprising any suitable conventional carrier, solvent, etc.

The amount of hydrocolloid to be given to the patient is preferably 5 - 10 g in half glass of water or of yoghurt ½ hour before the meal once or twice a day.

The present invention will now be illustrated with reference to the following Examples and accompanying Figs., without being restricted by them.

Example 1

The plant Portulaca Oleracea, was crushed for 2-3 minutes in an electric blender in the presence of ethanol in a ratio of 1:1 (wet plant:ethanol).

After crushing, the solid fraction was dried in an oven and extracted with acetone. The fraction rich with chlorophyll and

other dirty particles was deleted. Said fraction constitutes 0.5% of the weight of the dry plant. Thereafter, the remainder was extracted in a soxlet with a mixture of toluene:ethanol in a ratio of 1:2. The lipid fraction was thus removed. Said fraction comprises most of the lipids, the fats, the carotenoids, the phospholids and the wax. Said fraction constitutes 3% of the weight of the dry plant. Thereafter, the remainder was extracted with water. Said extraction may be performed at room temperature or at temperatures up to 100°C. The higher said temperature, the shorter the extraction time. In general, said extraction was performed in the course of 4 hours at 50°C.

The aqueous fraction was then centrifuged in order to remove the rest of the plant. Ethanol was then added to the aqueous fraction in a ratio of ethanol : aqueous solution 3 : 1. The water soluble hydrocolloid was precipitated.

After drying in an oven and in vacuum at room temperature said hydrocolloid constitutes about 3-5% of the weight of the plant. The product obtained is a white powder which is partly soluble in water. Some further part may be dispersed in water (see Table V).

Example 2

The hydrocolloid was dissolved in water at room temperature with magnetic stirring overnight. The oil was dripped into the solution and the stirring was performed in a homogenizer (9500 rpm). After 5 minutes all the oil had been dripped into the emulsion. Stirring was continued for another 10 minutes. Thereafter, if required, stirring is performed in a micro-fluidizer (3-4 turnovers).

By the above method the following emulsions were prepared:

a. 5% of tetra-decane was dropped into 95% aqueous solution

of 0.5% hydrocolloid with homogenization by ultraturrax (9500 rpm) and followed by micro-fluidization (4 - 5 turnovers). The emulsion obtained was stable more than a year. The size of the droplets was less than 5 μm.

b. In a similar manner as described in a. above an emulsion of 20% tetra-decane and 80% of a 0.5% aqueous hydrocolloid solution was prepared. The emulsion obtained was stable more than a fortnight.

c. In a similar manner as described in a. above an emulsion of 30 tetra-decane and 70% of a 0.9 aqueous solution was prepared. The emulsion obtained was stable more than a fortnight.

d. In a similar manner as described in a. above an emulsion of toluene (5%), of hydrocolloid (0.5 %0 and water (94.5%). was prepared. The emulsion obtained was so stable and transparent that nearly no drops could be seen.

Example 3

A serie of emulsions having different pH of the aqueous phase was prepared in a similar manner as described in Example 2. The concentration of the hydrocolloid was 0.5% of the emulsion. The oil was tetra-decane.

The hydrocolloid was dissolved, in the course of the night, in water with magnetic stirring. The pH was adjusted to 1.7, 3. 6.6, 9 and 11 by the addition of HCl or NaOH, respectively, without the addition of a buffer. The solution maintains the desired pH for an extended period of time. The oil was dripped into the solution with magnetic stirring.

The influence of the concentration of an electrolyte on the stability of the emulsion was also tested.

When the electrolyte was 0.015 M NaCl it was added to the

hydrocolloid solution before the oil was added.

The results are shown in Table VIII and in annexed Fig. 1.

ESI in accordance with the following table is:

ESI = the ratio between the percentage of the oil drops (in accordance with the volume) in the range of 2-10 μm a week after the day of preparation.

Table VIII

	1 Nacl=0.015M	2	3 No electrolyte
	ESI	pH	ESI
1	0.8695	1.7	
2	0.9208	3	0.9643
3	0.3968	6.6	0.78
4	0.347	9	0.588
5	0.2564	12	

As can be seen the stability of the emulsion is improved when the pH is decreased. The most stable emulsion is obtained around pH 3.

Example 4

The activity of the hydrocolloid prepared as described in Example 2 after being subject to ultracentrifugation (32000 rotations/30 minutes was tested. The results are given in Table IX.

Table IX

ESI	Kind of Hydrocolloid
0.396	Untreated hydrocolloid
0.945	Hydrocolloid after treatment by ultracentrifugation.

It can be seen that the hydrocolloid which was treated by ultracentrifugation was much more stable than the untreated

hydrocolloid

Example 5

In order to compare the activity of the hydrocolloid obtained from Portulaca Oleracea, i.e. that according to the present invention with emulsions comprising hydrocolloids of other origins there were prepared, in the same manner as described in Example 2 (the time of homogenisation was reduced from 15 to 5 minutes) emulsions comprising 0.5% hydrocolloid and 5% tetra-decane. The compared hydrocolloids were:

Arabic, Guar and Xanthan.

The results are shown in Table X.

ESI in accordance with the following table is:

$$ESI = \frac{\text{\% (of Volume) of drops 2 -10 } \mu m \text{ in a week}}{\text{\% (of Volume) of drops in the range of 2-10 } \mu m \text{ at the day of preparation}}$$

Table X

	Portulaca	Arabic	Guar	Xanthan
The size of the average drops, in volume, on the day of preparation	2.77 μm	2.575 μm	24.5 μm	16 μm
Stability of mix	stable	stable	stable	stable
ESI	0.91	0.62	phase sep.	phase sep.
flocculation	small	no	floc.	floc.

As can be seen the hydrocolloid according to the present invention gives the most stable emulsion in comparison with the other tested hydrocolloids.

Example 6

A serie of emulsions was prepared changing the dissolving

temperature of the hydrocolloid. The hydrocolloid was at first dissolved over the night at room temperature. Thereafter it was heated for 10 minutes to 50°, 70° and 90°C, respectively. Thereafter the emulsion was cooled slowly to room temperature. Then the emulsion was prepared as described in Example 2.

The results are shown in Table XI and in Fig. 2.

Table XI

	1	2
	Temp.	ESI
1	25	0.68
2	50	0.3968
3	70	0.37
4	90	0.3077

As can be seen the most stable emulsion is obtained when the hydrocolloid is not heated before the emulsion is prepared.

Example 7

A test was made to combine the hydrocolloid according to the present invention with a monomeric emulsifier.

A serie of emulsions was prepared in which the hydrocolloid and Tween 20 were prepared in 2 steps:

At the beginning a 0.3% emulsion was prepared in the following manner:

After it has been dissolved in the conventional manner, oil (tetra-decane) was dripped in, with homogenisation, into the solution. The homogenisation was performed for 15 minutes in a homogenisator. To the emulsion obtained was added Tween 20 in concentrations of 0.5 - 4%. The emulsion was stirred for 1 hour with a magnetic stirrer. Thereafter the Z-potential of the oil drops was measured. As the result of the measurement the composi-

tion of the drops was determined. With the increase of the concentration of the Tween 20 there occurs a gradual substitution of hydrocolloid by the Tween 20. An absolute substitution occurs in the ratio of the weight concentrations

Hydrocolloid/Tween 20 = 0.3wt%/4 wt%.

The concentration of Tween 20 required for the absolute substitution is relatively high to the proteins?, i.e. when the concentration of the Tween 20 is lower than 4% both emulsifiers are on the oil drops.

Example 8.

For checking the change of the blood pressure 5 - 10 g of the hydrocolloid in ½ a glass of water or yoghurt were given to 10 patients ½ hour before the meal. The tests were continued for a fortnight. The blood pressure was measured at the beginning of the test and at the end thereof. The blood pressure was reduced from 160 ± 20 /110 ± 20 to 125 ± 20 / 90 ± 20, respectively.

Example 9

6.4 mg/ml of sugar were dissolved in a in vitro/in vivo test on one side of a rat intestine. On the other side of the intestine only water was present. When a hydrocolloid emulsion was added to the sugar solution the amount thereof in the other side was only 1.7 mg/ml.

Example 10

The contents of the sugar in the blood was tested. 5 - 10 g of the hydrocolloid in ½ a glass of water or yoghurt in were given to 10 patients ½ hour before the meal. The tests were continued for a fortnight. The sugar content was measured at the beginning of the test and at the end thereof. At the end of the test the amount of glucose in the blood for those patients being fed with the

hydrocolloid was 135 mg glucose/dl blood, whereas the amount in the blood of untreated patients was 180 mg glucose/dl blood.

Claims

1. A hydrocolloid obtained by extraction from Portulaca Oleracea having the composition as shown in Table I in which the sugar contents in the fibers are as shown in Table II and the composition of the ash portion is as given in Table III.
2. A hydrocolloid according to Claim 1 having the characteristic features as given in Tables IV - VII.
3. An emulsion comprising a hydrocolloid according to Claim 1 or 2.
4. An emulsion according to Claim 3 being a tetra-decane in an aqueous solution of the hydrocolloid.
5. An emulsion as defined in Claim 3, substantially as herein described with reference to Examples 2 to 7.
6. A pharmaceutical composition preparation comprising as active ingredient a hydrocolloid according to Claim 1 or 2.
7. A pharmaceutical composition according to Claim 6 being part of an emulsion according to any of Claims 3 to 5.
8. A pharmaceutical composition according to Claim 6 or 7 for reducing the sugar content in the blood.
9. A pharmaceutical composition according to Claim 6 or 7 for reducing the blood pressure.

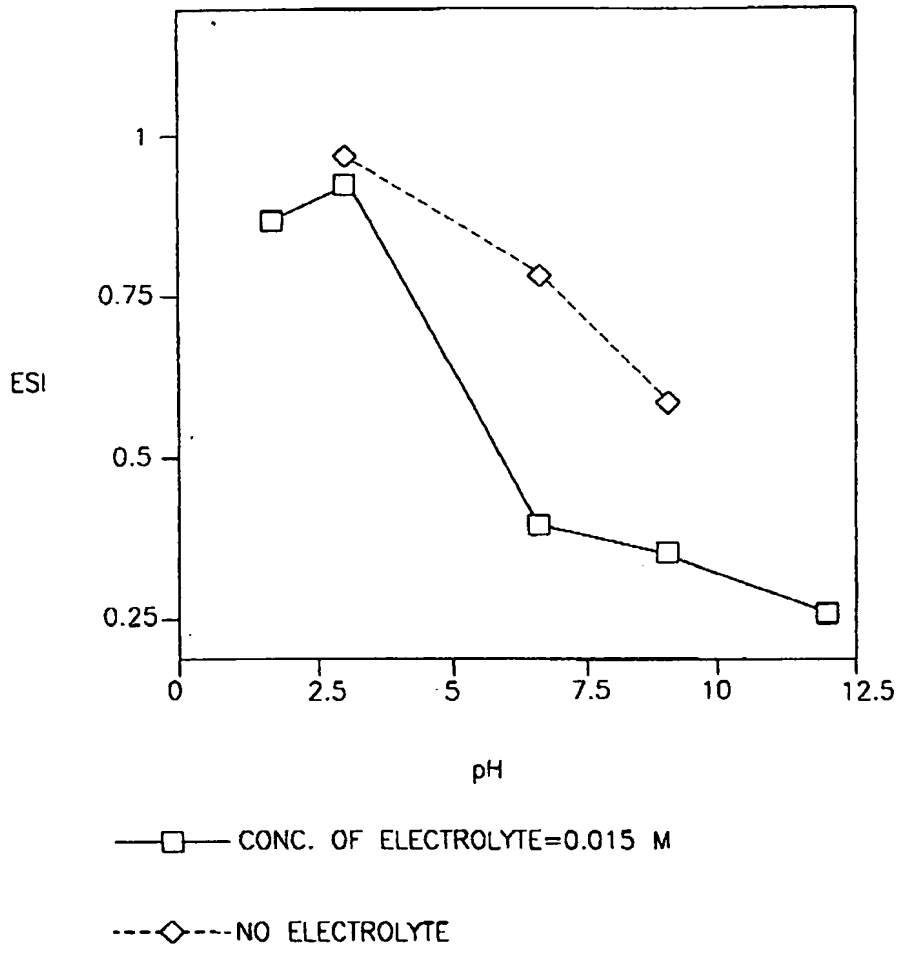
1
0.75
0.5
0.25
ESI
0
2.5
5
7.5
10
12.5
pH
CONC. OF ELECTROLYTE=0.015 M
NO ELECTROLYTE

FIG.1

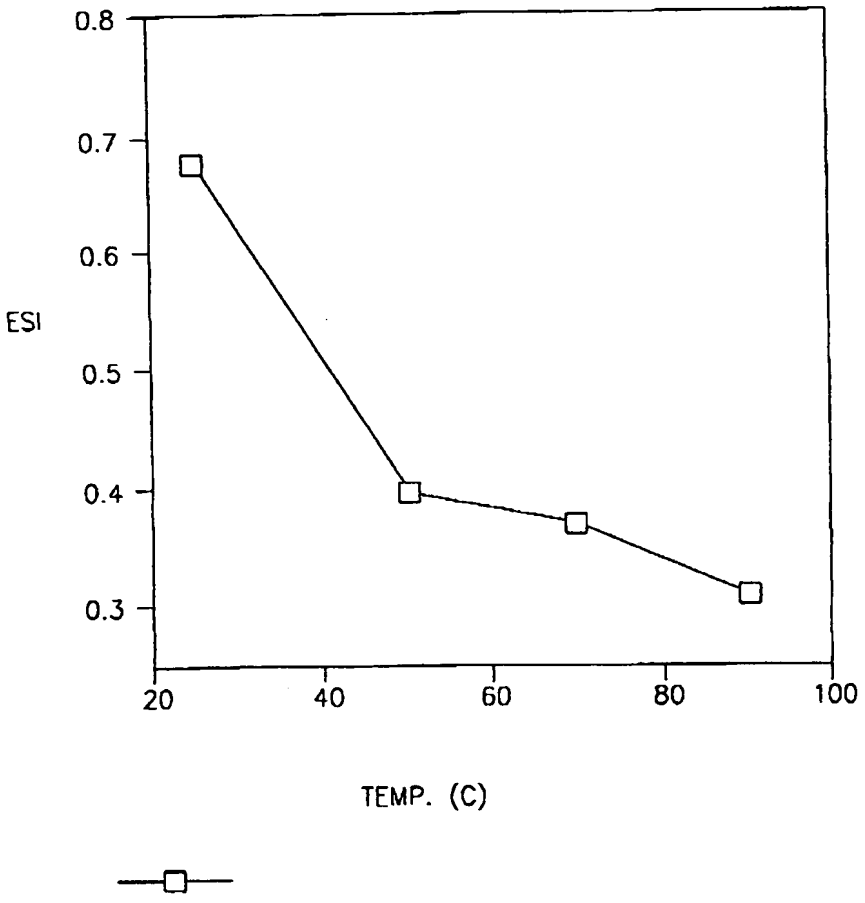

FIG.2

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
6 January 2000 (06.01.2000)

PCT

(10) International Publication Number
WO 00/000211 A3

(51) International Patent Classification[7]: A61K 35/78

(21) International Application Number: PCT/IL99/00355

(22) International Filing Date: 28 June 1999 (28.06.1999)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
125138 29 June 1998 (29.06.1998) IL

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(81) Designated States *(national)*: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW.

(84) Designated States *(regional)*: ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published:
— *with international search report*

(88) Date of publication of the international search report:
17 October 2002

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

WO 00/000211 A3

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(57) Abstract: The present invention relates to a hydrocolloid obtained by extraction from Portulaca Oleracea having the composition as shown in Table I in which the sugar contents in the fibers are as shown in Table II and the composition of the ash portion is as given in Table III.

INTERNATIONAL SEARCH REPORT

International Application No
PCT/IL 99/00355

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 A61K35/78

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

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C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 98 24457 A (EASTWOOD BIOMEDICAL RESEARCH I) 11 June 1998 (1998-06-11) ---	
A	GUIDO E. WENZEL ET AL.: "THE VISCOUS MUCILAGE FROM THE WEED PORTULACA OLERACEA, L." APPLIED BIOCHEMISTRY AND BIOTECHNOLOGY, vol. 24/25, 1990, pages 341-353, XP002118259 CLIFTON, NJ, US ISSN: 0273-2289 cited in the application --- -/--	

[X] Further documents are listed in the continuation of box C.

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Date of the actual completion of the international search	Date of mailing of the international search report
8 October 1999	25/10/1999

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Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

International Application No
PCT/IL 99/00355

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication,where appropriate, of the relevant passages	Relevant to claim No.
A	O. PARRY ET AL.: "EFFECT OF AN AQUEOUS EXTRACT OF PORTULACA OLERACEA LEAVES ON SMOOTH MUSCLE AND RAT BLOOD PRESSURE." JOURNAL OF ETHNOPHARMACOLOGY., vol. 22, no. 1, January 1998 (1998-01), pages 33-44, XP002118260 ELSEVIER SCIENTIFIC PUBLISHERS LTD., IE ISSN: 0378-8741	

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/IL 99/00355

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		US 5869060 A	09-02-1999
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